

This Page Is Inserted by IFW Operations
and is not a part of the Official Record

BEST AVAILABLE IMAGES

Defective images within this document are accurate representations of the original documents submitted by the applicant.

Defects in the images may include (but are not limited to):

- BLACK BORDERS
- TEXT CUT OFF AT TOP, BOTTOM OR SIDES
- FADED TEXT
- ILLEGIBLE TEXT
- SKEWED/SLANTED IMAGES
- COLORED PHOTOS
- BLACK OR VERY BLACK AND WHITE DARK PHOTOS
- GRAY SCALE DOCUMENTS

IMAGES ARE BEST AVAILABLE COPY.

**As rescanning documents *will not* correct images,
please do not report the images to the
Image Problem Mailbox.**

PATENT SPECIFICATION

(11) 1 261 660

1 261 660

NO DRAWINGS

- (21) Application No. 28940/70 (22) Filed 15 June 1970
 (31) Convention Application No. 835 307 (32) Filed 18 June 1969
 (31) Convention Application No. 22076 (32) Filed 23 March 1970 in
 (33) United States of America (US)
 (45) Complete Specification published 26 Jan. 1972
 (51) International Classification C 07 c 109/04; A 61 k 27/00 // C 07 c 127/14



- (52) Index at acceptance
 C2C 220 227 22Y 29X 29Y 30Y 327 341 34Y 364 365
 366 367 368 36Y 626 657 70X 743 790 79Y KD
 MF

ASB 385 387 38Y 392 421 42Y 431 432 43Y 482 48Y
 586 58Y 77Y

- (72) Inventors SANDOR KARADY, SEEMON HAYDEN PINES,
 MANUEL GUING LY and MEYER SLETZINGER

(54) α -HYDRAZINO ACIDS

- (71) We, MERCK & CO INC, a corporation duly organised and existing under the laws of the State of New Jersey, United States of America, of Rahway, New Jersey, United States of America, do hereby declare the invention, for which we pray that a patent may be granted to us, and the method by which it is to be performed, to be particularly described in and by the following statement: —
- 10 Certain α - hydrazino - α - substituted - β - (3,4 - dihydroxyphenyl) - propionic acids and their esters and salts form part of the invention of our prior Patent No 940596. It is known that racemates of these compounds are very potent decarboxylase inhibitors in mammals: see Sletzinger et al "Journal of Medicinal Chemistry", Volume 6, page 101 (1963) and Porter et al "Biochemical Pharmacology", Volume 11, page 1067 (November 1962). Such compounds have found widespread use in the pharmaceutical field. It has now been found that the *D* isomer of the compound is essentially inactive and may even be antagonistic to the action of the *L* form thereby reducing its potency. Thus, in some tests it appears that the *L* form of the compound is the only active form and the *D* form is completely inactive and in other tests it appears that the *D* form will actually detract from the action of the *L* form.
- 20 The new compounds of the present invention are *L* - α - hydrazino - β - (3,4-dihydroxyphenyl)propionic acid substantially free of the *D* isomer, *L* - α - hydrazino - α - (C₁₋₃ alkyl) - β - (3,4 - dihydroxyphenyl) - propionic acids, particularly *L* - α - hydrazino - α - methyl - β - (3,4 - dihydroxyphenyl) - propionic acid and *L* - α - hydrazino - ethyl - β - (3,4 - dihydroxyphenyl)propionic acid, substantially free of the *D* isomer, and their C₁₋₃

alkyl esters, and pharmaceutically acceptable salts of the said acids and esters. Compounds of the present invention have been found to be much more potent decarboxylase inhibitors than were heretofore available to the medical profession. Such compounds have also been found to inhibit histidine decarboxylase, and thus to show promise of use as anti-histaminics as well.

The inhibition of mammalian decarboxylase is an important part of the physiological action of many types of drugs. For example, it has recently been proposed to use *L*-dopa in the treatment of Parkinson's disease. However, *L*-dopa is utilized both in the brain and the peripheral parts of the body and it is desired that it be utilized only in the brain. Hydrazino compounds of the present invention have been found not to pass the blood-brain barrier and hence they inhibit decarboxylase only in the peripheral parts of the body. Thus, when *L*-dopa is used in conjunction with the hydrazino compounds of the present invention, the decarboxylase of *L*-dopa is inhibited only in the peripheral parts of the body making more of it available to the brain. The net result is that much less *L*-dopa is required for effective medication.

The inhibition of decarboxylase is also of importance in the treatment of certain disorders of the colon. In some persons, the cells in the intestines and perhaps elsewhere develop overactivity in the production of serotonin from 5-hydroxytryptophane. The result of such overabundance of serotonin is constant flushing of the colon and evacuation of the bowels. Further, unless this condition is controlled, it can develop into much more serious trouble. Decarboxylase inhibitors prevent the formation of the serotonin and therefore control such

C. Preparation of *L* - α - methyl - β - (3,4-dihydroxyphenyl) - α - hydrazino propionic acid

5 A mixture of the *L* - α - (3,4 - dimethoxybenzyl) - α - hydrazino propionic acid (10 g.) and concentrated hydrochloric acid (150 ml.) is heated in a sealed tube at 120°C. for 2 hours. The reaction mixture is evaporated to dryness *in vacuo* and the product is leached out with ethanol. The hydrazino acid is precipitated by the addition of diethylamine to

pH 6.4. The precipitate is filtered off, washed with ethanol and dried, affording 6.5 g of *L* - α - methyl - β - (3,4 - dihydroxyphenyl) - α - hydrazino propionic acid (73%). Recrystallization from water (containing a small amount of sodium bisulfite) yields analytically pure material; m.p. 208° dec.

AClCl₃
[α] -25.4°
546

20 Anal. calcd. for C₁₀H₁₄N₂O₄ · H₂O:
Found:

C, 49.17; H, 6.60; N, 11.47.
C, 49.13; H, 6.74; N, 11.19.

EXAMPLE 2

A. Preparation of *L* - 4 - (3,4 - dimethoxybenzyl) - 4 - ethylhydantoic acid

25 The procedure of Example 1A is repeated using *L* - α - amino - α - (3,4 - dihydroxybenzyl)butyric acid rather than the alanine

compound. The *L* - (4 - (3,4 - dimethoxybenzyl) - 4 - ethylhydantoic acid is obtained in a yield of 70%. The product is recrystallized from ethanol-water to give a pure product having a melting point of 218—220°C.

Anal. calcd. for C₁₄H₂₀O₆N₂:
Found:

C, 56.74; H, 6.80; N, 9.45.
C, 56.71; H, 6.88; N, 9.53.

35 B. Preparation of *L* - α - (3,4 - dimethoxybenzyl) - α - hydrazinobutyric acid

The procedure of Example 1B is repeated using the *L* - 4 - (3,4 - dimethoxybenzyl) - 4 - ethylhydantoic acid to obtain a 53% yield of

L - α - (3,4 - dimethoxybenzyl) - α - hydrazinobutyric acid. An analytical sample is recrystallized from ethanol-water to give a pure product having a melting point of 215—220°C.

45 Anal. calcd. for C₁₅H₂₀O₄N₂:
Found:

C, 58.19; H, 7.51; N, 10.44.
C, 58.16; H, 7.60; N, 10.40.

C. Preparation of *L* - α - hydrazino - α - ethyl - β - (3,4 - dihydroxyphenyl)propionic acid

50 The procedure of Example 1C is repeated using *L* - α - (3,4 - dimethoxybenzyl) - α - hydrazinobutyric acid to give a 90% yield of *L* - α - hydrazino - α - ethyl - β - (3,4-

dihydroxyphenyl)propionic acid which, when recrystallized from water-isopropanol, has a melting point of 209—212°C.

H₂O
[α] -15.2 C=1
D

Anal. calcd. for C₁₁H₁₆O₄N₂:
Found:

C, 54.99; H, 6.71; N, 11.66.
C, 55.02; H, 6.70; N, 11.65.

EXAMPLE 3

60 The procedure of Example 1A, 1B and 1C is repeated using *L* - β - (3,4 - dihydroxyphenyl)alanine as the starting material. The resulting product is *L* - α - hydrazino - β - (3,4 - dihydroxyphenyl)propionic acid.

the indicated dose of *L* - α - hydrazino - α - methyl - β - (3,4 - dihydroxyphenyl)propionic acid orally in a solution or suspension in water. The animals are decapitated 90 minutes later.

The brains are removed and pooled in groups of seven. Three separate pools are used for each drug treatment and the values obtained are averaged.

The brains are homogenized with 0.4 N perchloric acid, 9 ml. per gram of tissue. Catecholamines and catecholaminoacids are adsorbed onto and then eluted from alumina. The dopa and dopamine are separated by chromatography utilizing a column containing

EXAMPLE 4

Testing for decarboxylase inhibition in mammals

70 Female albino mice weighing between 18 to 22 g. each are used. The animals are administered 80 mg./kg. of *L*-dopa (*L* - 3,4-dihydroxyphenylalanine) in combination with

TABLE 2

Comparison of the effect of the *D* and *L* isomers of α -hydrazino- α -methyl- β -(3,4-dihydroxyphenyl) propionic acid upon *L*-dopa antagonism of reserpine-induced suppression of locomotion and ptosis

Pretreatment ^a	Dose (mg/kg) p.o.)	Reserpine-induced Suppression of Locomotion	Reserpine-induced Suppression of Ptosis
		No. mice protected No. mice tested	No. mice protected No. mice tested
Methocel	—	1/10	1/10
<i>D</i> isomer + Methocel	5.0	1/10	1/10
	25.0	2/10	2/10
	125.0	2/10	3/10
<i>L</i> isomer + Methocel	0.2	3/10	2/10
	1.0	5/10	7/10
	5.0 ^b	8/10	9/10
ED ₅₀ ^c		0.86 mg/kg	0.56 mg/kg

a — One hour prior to *L*-dopa 150 mg/kg i.p.

b — This dose of *L*- α -hydrazino- α -methyl- β -(3,4-dihydroxyphenyl) propionic acid was inactive as a reserpine antagonist when administered prior to Methocel.

c — Estimated dose of α -hydrazino- α -methyl- β -(3,4-dihydroxyphenyl) propionic acid, when given in combination with *L*-dopa (150 mg/kg i.p.), necessary to antagonize these effects of reserpine in 50% of the mice.

WHAT WE CLAIM IS:—

1. *L* - α - hydrazino - β - (3,4 - dihydroxyphenyl)propionic acid substantially free of the *D*-isomer.
- 5 2. *L* - α - hydrazino - α - C₁₋₅ alkyl) - β - (3,4 - dihydroxyphenyl)propionic acids substantially free of the *D*-isomer.
3. *L* - α - hydrazino - α - methyl - β - (3,4 - dihydroxyphenyl)propionic acid substantially free of the *D*-isomer.
- 10 4. *L* - α - hydrazino - α - ethyl - β - (3,4 - dihydroxyphenyl)propionic acid substantially free of the *D*-isomer.
5. The C₁₋₅ alkyl esters of a compound claimed in any one of claims 1—4.
- 15 6. A pharmaceutically acceptable salt of a compound claimed in any one of claims 1—5.
7. The preparation of a compound as claimed in any one of claims 1—6 by a process substantially as hereinbefore described in any appropriate Example.
- 20 8. A compound as claimed in any one of claims 1—6 when made by a preparation as claimed in claim 7 or its obvious chemical equivalent.
9. A method of inhibiting mammalian decarboxylase that comprises administering to a non-human mammal from 0.05 to 100 mg/kg per day of a compound as claimed in any one of claims 1—6 and 8.
10. A pharmaceutical composition comprising an inert pharmaceutical carrier and from about 5 mg to about 15 g of a compound as claimed in any one of claims 1—6 and 8.
11. A composition as claimed in claim 10 in the form of a pill, tablet, capsule, syrup, elixir or injectable preparation.

For the Applicants:
D. YOUNG & CO.,
Chartered Patent Agents,
9 Staple Inn, London, W.C.1.

Printed for Her Majesty's Stationery Office by the Courier Press, Leamington Spa, 1972.
Published by the Patent Office, 25 Southampton Buildings, London, WC2A 1AY, from which copies may be obtained.